



SEQUENCE LISTING

<110> WOULFE, SUSAN L.
JAIN, RITA
BURR, AIMEE

<120> ENGINEERED FAB' FRAGMENT ANTI-TUMOR
NECROSIS FACTOR ALPHA IN COMBINATION WITH DISEASE MODIFYING
ANTI-RHEUMATIC DRUGS

<130> 122294-1010

<140> US/10/728,420

<141> 2003-12-05

<150> US 60/431,053

<151> 2002-12-05

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<223> Mouse monoclonal antibody hTNF40/human hybrid CDRH2

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Trp Ile Asn Thr Tyr Ile Gly Glu Pro Ile Tyr Ala Asp Ser Val Lys
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<223> Mouse monoclonal antibody hTNF40 CDRH3

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Gly Tyr Arg Ser Tyr Ala Met Asp Tyr
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 Ser Ala Ser Phe Leu Tyr Ser
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Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly	
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gac cgg gtc acc atc act tgt aaa gcc agt cag aac gta ggt act aac	96
Asp Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Asn Val Gly Thr Asn	
20 25 30	

gta gcc tgg tat cag caa aaa cca ggt aaa gcc cca aaa gcc ctc atc	144
Val Ala Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Ala Leu Ile	
35 40 45	

tac agt gcc tct ttc ctc tat agt ggt gta cca tac agg ttc agc gga	192
Tyr Ser Ala Ser Phe Leu Tyr Ser Gly Val Pro Tyr Arg Phe Ser Gly	
50 55 60	

tcc ggt agt ggt act gat ttc acc ctc acg atc agt agc ctc cag cca	240
Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro	
65 70 75 80	

gaa gat ttc gcc act tat tac tgt caa cag tat aac atc tac cca ctc	288
Glu Asp Phe Ala Thr Tyr Tyr Cys Gln Gln Tyr Asn Ile Tyr Pro Leu	
85 90 95	

aca ttc ggt cag ggt act aaa gta gaa atc aaa	321
Thr Phe Gly Gln Gly Thr Lys Val Glu Ile Lys	
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<222> (1)...(354)

<223> Grafted Heavy Chain for Modified Fab

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gag gtt cag ctg gtc gag tca gga ggc ggt ctc gtg cag cct ggc gga	48
Glu Val Gln Leu Val Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly	
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tca ctg aga ttg tcc tgt gct gca tct ggt tac gtc ttc aca gac tat	96
Ser Leu Arg Leu Ser Cys Ala Ala Ser Gly Tyr Val Phe Thr Asp Tyr	

20

25

30

gga atg aat tgg gtt aga cag gcc ccg gga aag ggc ctg gaa tgg atg 144
Gly Met Asn Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Met
35 40 45

ggt tgg att aat act tac att gga gag cct att tat gct gac agc gtc 192
Gly Trp Ile Asn Thr Tyr Ile Gly Glu Pro Ile Tyr Ala Asp Ser Val
50 55 60

aag ggc aga ttc acg ttc tct cta gac aca tcc aag tca aca gca tac 240
Lys Gly Arg Phe Thr Phe Ser Leu Asp Thr Ser Lys Ser Thr Ala Tyr
65 70 75 80

ctc caa atg aat agc ctg aga gca gag gac acc gca gtg tac tat tgt 288
Leu Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys
85 90 95

gct aga gga tac aga tct tat gcc atg gac tac tgg ggc cag ggt acc 336
Ala Arg Gly Tyr Arg Ser Tyr Ala Met Asp Tyr Trp Gly Gln Gly Thr
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Leu Val Thr Val Ser Ser
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<223> human group 1 consensus framework L1

<400> 83

Asp	Ile	Gln	Met	Thr	Gln	Ser	Pro	Ser	Ser	Leu	Ser	Ala	Ser	Val	Gly
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Asp	Arg	Val	Thr	Ile	Thr	Cys									
			20												

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<212> PRT

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<223> Human hTNF40 framework L1

<400> 84

Asp	Ile	Val	Met	Thr	Gln	Ser	Gln	Lys	Phe	Met	Ser	Thr	Ser	Val	Gly
1				5				10						15	
Asp	Arg	Val	Ser	Val	Thr	Cys									
			20												

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<212> PRT

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Trp	Tyr	Gln	Gln	Lys	Pro	Gly	Lys	Ala	Pro	Lys	Leu	Leu	Ile	Tyr
1				5				10					15	

<210> 86

<211> 15

<212> PRT

<213> Artificial Sequence

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<223> Human hTNF40 framework L2

Sequence Listing 015181-1 US rev 09-14-04.txt

<400> 86

Trp Tyr Gln Gln Lys Pro Gly Gln Ser Pro Lys Ala Leu Ile Tyr
1 5 10 15

<210> 87

<211> 32

<212> PRT

<213> Artificial Sequence

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<223> human group 1 consensus framework L3

<400> 87

Gly Val Pro Ser Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr
1 5 10 15
Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp Phe Ala Thr Tyr Tyr Cys
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<210> 88

<211> 32

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<223> Human hTNF40 framework L3

<400> 88

Gly Val Pro Tyr Arg Phe Thr Gly Ser Gly Ser Gly Thr Asp Phe Thr
1 5 10 15
Leu Thr Ile Ser Thr Val Gln Ser Glu Asp Leu Ala Glu Tyr Phe Cys
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<210> 89

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<223> human group 1 consensus framework L4

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Phe Gly Gln Gly Thr Lys Val Glu Ile Lys Arg
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<210> 90

<211> 11

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<223> Human hTNF40 framework L4

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Phe Gly Ala Gly Thr Lys Leu Glu Leu Lys Arg
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Sequence Listing 015181-1 US rev 09-14-04.txt

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<212> PRT

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<223> human group 1 consensus framework H1

<400> 91

Gln Val Gln Leu Val Gln Ser Gly Ala Glu Val Lys Lys Pro Gly Ala
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 Ser Val Lys Val Ser Cys Lys Ala Ser Gly Tyr Thr Phe Thr
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<210> 92

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<223> Human hTNF40 framework H1

<400> 92

Gln Ile Gln Leu Val Gln Ser Gly Pro Glu Leu Lys Lys Pro Gly Glu
 1 5 10 15
 Thr Val Lys Ile Ser Cys Lys Ala Ser Gly Tyr Val Phe Thr
 20 25 30

<210> 93

<211> 14

<212> PRT

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<223> human group 1 consensus framework H2

<400> 93

Trp Val Arg Gln Ala Pro Gly Gln Gly Leu Glu Trp Met Gly
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<210> 94

<211> 14

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<223> Human hTNF40 framework H2

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Trp Val Lys Gln Ala Pro Gly Lys Ala Phe Lys Trp Met Gly
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Sequence Listing 015181-1 US rev 09-14-04.txt

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Arg Val Thr Ile Thr Arg Asp Thr Ser Thr Ser Thr Ala Tyr Met Glu
 1 5 10 15
 Leu Ser Ser Leu Arg Ser Glu Asp Thr Ala Val Tyr Tyr Cys Ala Arg
 20 25 30

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<400> 96

Arg Phe Ala Phe Ser Leu Glu Thr Ser Ala Ser Thr Ala Phe Leu Gln
 1 5 10 15
 Ile Asn Asn Leu Lys Asn Glu Asp Thr Ala Thr Tyr Phe Cys Ala Arg
 20 25 30

<210> 97

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<400> 97

Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser
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<210> 98

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<223> human group 1 consensus framework H4

<400> 98

Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser
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<211> 324

<212> DNA

<213> murine

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<222> (1)...(324)

<223> mouse hTNF40 light chain variable domain

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gac att gtg atg acc cag tct caa aaa ttc atg tcc aca tca gta gga 48
 Asp Ile Val Met Thr Gln Ser Gln Lys Phe Met Ser Thr Ser Val Gly
 1 5 10 15

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gac agg gtc agc gtc acc tgc aag gcc agt cag aat gtg ggt act aat 96
Asp Arg Val Ser Val Thr Cys Lys Ala Ser Gln Asn Val Gly Thr Asn
      20                25                30

gta gcc tgg tat caa cag aaa cca gga caa tct cct aaa gca ctg aat 144
Val Ala Trp Tyr Gln Gln Lys Pro Gly Gln Ser Pro Lys Ala Leu Asn
      35                40                45

tac tcg gca tcc ttc cta tat agt gga gtc cct tat cgc ttc aca ggc 192
Tyr Ser Ala Ser Phe Leu Tyr Ser Gly Val Pro Tyr Arg Phe Thr Gly
      50                55                60

agt gga tct ggg aca gat ttc act ctc acc atc agc act gtg cag tct 240
Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Thr Val Gln Ser
      65                70                75

gaa gac ttg gca gag tat ttc tgt cag caa tat aac atc tat cct ctc 288
Glu Asp Leu Ala Glu Tyr Phe Cys Gln Gln Tyr Asn Ile Tyr Pro Leu
      85                90                95

acg ttc ggt gct ggg acc aag ctg gag ctg aaa cgt 324
Thr Phe Gly Ala Gly Thr Lys Leu Glu Leu Lys Arg
      100                105

<210> 100
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<223> mouse hTNF40 heavy chain variable domain

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cag atc cag ttg gtg cag tct gga cct gag ctg aag aag cct gga gag 48
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      1                5                10                15

aca gtc aag atc tcc tgc aag gct tct gga tat gtt ttc aca gac tat 96
Thr Val Lys Ile Ser Cys Lys Ala Ser Gly Tyr Val Phe Thr Asp Tyr
      20                25                30

gga atg aat tgg gtg aag cag gct cca gga aag gct ttc aag tgg atg 144
Gly Met Asn Trp Val Lys Gln Ala Pro Gly Lys Ala Phe Lys Trp Met
      35                40                45

ggc tgg ata aac acc tac att gga gag cca ata tat gtt gat gac ttc 192
Gly Trp Ile Asn Thr Tyr Ile Gly Glu Pro Ile Tyr Val Asp Asp Phe
      50                55                60

aag gga cga ttt gcc ttc tct ttg gaa acc tct gcc agc act gcc ttt 240
Lys Gly Arg Phe Ala Phe Ser Leu Glu Thr Ser Ala Ser Thr Ala Phe
      65                70                75

ttg cag atc aac aac ctc aaa aat gag gac acg gct aca tat ttc tgt 288
Leu Gln Ile Asn Asn Leu Lys Asn Glu Asp Thr Ala Thr Tyr Phe Cys
      85                90                95

gca aga ggt tac cgg tcc tat gct atg gac tac tgg ggt caa gga acc 336
Ala Arg Gly Tyr Arg Ser Tyr Ala Met Asp Tyr Trp Gly Gln Gly Thr
      100                105                110

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tca gtc acc gtc tct tca
Ser Val Thr Val Ser Ser
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354

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<222> (29)...(67)
<223> Ompa oligonucleotide adaptor

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tcgagttcta gataacgagg cgtaaaaaat gaaaaagaca gctatcgcaa ttgcagtggc 60
cttggtctg acgtacgagt cagg 84

<210> 102
<211> 67
<212> DNA
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<222> (2)...(40)
<223> IGS cassette-1

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g agc tca cca gta aca aaa agt ttt aat aga gga gag tgt ta atg aag 48
Ser Ser Pro Val Thr Lys Ser Phe Asn Arg Gly Glu Cys Met Lys
1 5 10 15

aag act gct ata gca att g 67
Lys Thr Ala Ile Ala Ile
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<400> 103
g agc tca cca gta aca aaa agt ttt aat aga ggg gag tgt taa 43

Sequence Listing 015181-1 US rev 09-14-04.txt

Ser Ser Pro Val Thr Lys Ser Phe Asn Arg Gly Glu Cys *
 1 5 10

a atg aag aag act gct ata gca att g 69
 Met Lys Lys Thr Ala Ile Ala Ile
 15 20

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 Ser Ser Pro Val Thr Lys Ser Phe Asn Arg Gly Glu Cys *
 1 5 10

ggaggaaaaa aaa atg aag aaa act gct ata gca att g 81
 Met Lys Lys Thr Ala Ile Ala Ile
 15 20

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 Met Lys Lys Thr Ala Ile Ala Ile
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<210> 106
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Sequence Listing 015181-1 US rev 09-14-04.txt

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<223> human group 3 consensus framework H1

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Glu Val Gln Leu Val Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly
1 5 10 15

Ser Leu Arg Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Ser
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<223> human group 3 consensus framework H2

<400> 107

Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Val Ser
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<210> 108

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<223> human group 3 consensus framework H3

<400> 108

Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Leu Tyr Leu Gln
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Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys Ala Arg
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Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser
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<223> Synthetic CDP870 Light chain mature protein sequence

<400> 113

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Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly
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Asp Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Asn Val Gly Thr Asn
 20           25           30
Val Ala Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Ala Leu Ile
 35           40           45
Tyr Ser Ala Ser Phe Leu Tyr Ser Gly Val Pro Tyr Arg Phe Ser Gly
 50           55           60
Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro
 65           70           75           80
Glu Asp Phe Ala Thr Tyr Tyr Cys Gln Gln Tyr Asn Ile Tyr Pro Leu
 85           90           95
Thr Phe Gly Gln Gly Thr Lys Val Glu Ile Lys Arg Thr Val Ala Ala
100           105           110
Pro Ser Val Phe Ile Phe Pro Pro Ser Asp Glu Gln Leu Lys Ser Gly
115           120           125
Thr Ala Ser Val Val Cys Leu Leu Asn Asn Phe Tyr Pro Arg Glu Ala
130           135           140
Lys Val Gln Trp Lys Val Asp Asn Ala Leu Gln Ser Gly Asn Ser Gln
145           150           155           160
Glu Ser Val Thr Glu Gln Asp Ser Lys Asp Ser Thr Tyr Ser Leu Ser
165           170           175
Ser Thr Leu Thr Leu Ser Lys Ala Asp Tyr Glu Lys His Lys Val Tyr
180           185           190
Ala Cys Glu Val Thr His Gln Gly Leu Ser Ser Pro Val Thr Lys Ser
195           200           205
Phe Asn Arg Gly Glu Cys
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<210> 114

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<210> 115

<211> 229

<212> PRT

<213> Artificial Sequence

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<223> Synthetic CDP870 Heavy chain mature protein sequence

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Glu Val Gln Leu Val Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly
 1           5           10           15

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Sequence Listing 015181-1 US rev 09-14-04.txt

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Ser Leu Arg Leu Ser Cys Ala Ala Ser Gly Tyr Val Phe Thr Asp Tyr
 20      25      30
Gly Met Asn Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Met

      35      40      45
Gly Trp Ile Asn Thr Tyr Ile Gly Glu Pro Ile Tyr Ala Asp Ser Val
 50      55      60
Lys Gly Arg Phe Thr Phe Ser Leu Asp Thr Ser Lys Ser Thr Ala Tyr
 65      70      75      80
Leu Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys
      85      90      95
Ala Arg Gly Tyr Arg Ser Tyr Ala Met Asp Tyr Trp Gly Gln Gly Thr
100      105      110
Leu Val Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser Val Phe Pro
115      120      125
Leu Ala Pro Ser Ser Lys Ser Thr Ser Gly Gly Thr Ala Ala Leu Gly
130      135      140
Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val Ser Trp Asn
145      150      155      160
Ser Gly Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala Val Leu Gln
      165      170      175
Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val Pro Ser Ser
180      185      190
Ser Leu Gly Thr Gln Thr Tyr Ile Cys Asn Val Asn His Lys Pro Ser
195      200      205
Asn Thr Lys Val Asp Lys Lys Val Glu Pro Lys Ser Cys Asp Lys Thr
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His Thr Cys Ala Ala
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<210> 116

<211> 1477

<212> DNA

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<223> synthetic CDP870 nucleic acid sequence (sense strand)

<400> 116

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accatcactt gtaaagccag tcagaacgta ggtactaacg tagcctggta tcagcaaaaa 180
ccaggtaaag ccccaaaagc cctcatctac agtgcctctt tcctctatag tgggtgtacca 240
tacaggttca gcggatccgg tagtggtact gatttcaccc tcacgactag tagcctccag 300
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Sequence Listing 015181-1 US rev 09-14-04.txt

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